Grinding of lyophilized mycelial pads

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Abstract
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the use of tests done in 4" test tubes closed with Oxoid Caps. Both are described in Catcheside 1960 Proc. Roy. Soc. London B153:179; more detail of the plate tests is given in Ahmad and Catcheside 1960 Heredity 15:55. For the tests in tubes we use baskets holding 64 tubes in 8 rows of 8 each. Each basket is labelled in a standard fashion with a code number corresponding to the protocol of the matrix to be set up. Drops of conidial suspension are added to each by means of Pasteur pipettes. The medium contains agar, not sloped, so that the conidial mixture sits on the top and is easily visible on inspection. Daily records are kept on record sheets, for up to 10 to 14 days. Beyond this time the medium tends to dry out too much, so concentrating the constituents. The concentration of the medium, as well as other factors such as the temperature of incubation, affects the ability to grow.

Crossing methods. We use the standard Westergaard and Mitchell formula in 6" tubes. A piece of folded filter paper is inserted into the medium, which contains agar and is sloped. The female parent is allowed to grow first and when abundant protoperithecia are seen to have been formed, the conidial parent is added. For conidiation, quite dense suspensions of conidia are made in 2 ml. of sterile distilled water and this suspension is then added to the slopes containing the female parent, after clearing out any excessive conidial growth that there may be. The tube is rotated between the hands to distribute the conidia and, after a time, the excess liquid is decanted. - - - Department of Genetics, John Curtin School of Medical Research, Australian National University, Canberra, A. C. T., Australia.

Fankhauser, D. B. Grinding of lyophilized mycelial pads. The grinding with mortar and pestle of lyophilized mycelial pads grown in 125 ml flasks (containing 50 ml of medium) has proven very difficult and time consuming. A much quicker and more effective method is as follows: The lyophilized pad is placed in an 18 x 150 mm test tube and 'chopped' into small pieces with two stainless steel spatulas (8" long with a flat end 2" x 1/4" ). With the spatulas still in the tube, and with the top of the tube held firmly, it is placed inside the cup of a Vortex Jr. Mixer and agitated for 15 to 20 seconds, giving a semi-fine to fine powder according to the length of agitation. A tube will occasionally chip at the top, but this can be minimized by checking for cracked tubes beforehand, and by holding the tube at the top. 20 x 150 mm tubes should not be used because they break too easily. The tube should not be pressed down with any more force than is necessary to hold it in the cup because contact with the screw at the cup base will scratch, and eventually break, the bottom of the tube. In three months, we have never had a tube disintegrate.

This method will grind up to 400 mg of powder, yielding as fine a powder as desired. The enzymes, tryptophan synthetase and indole glycerol phosphate synthetase, are extracted as completely from these powders as from those prepared by use of mortar and pestle. - - - Department of Microbiology, University of Cincinnati, College of Medicine, Cincinnati, Ohio.

Kilbey, B. J. The detection of irreparable mutants in Neurospora. The heterokaryon system used by Atwood and Mukai (1953 Proc. Natl. Acad. Sci. U. S. 39:1027) for the detection of irreparable mutants in Neurospora is open to two main criticisms: first, the heterokaryotic component in which irreparable mutants are scored carries the amyecelial and methionineless genetic markers, and, second, the tests for reparability are made with medium containing sorbose. Both the genetic background and the plating environment are probably unfavorable for the detection of reparable mutants (Horowitz 1963 NN#3:5).

In an attempt to obviate these criticisms, an entirely new heterokaryon has been prepared. Both the components of the heterokaryon have been derived from the K3/17 strain of Kalmark (Kalmark and Kilbey 1962 Zetl. für Vererbungslehre 93:356). This strain carries a complex of colonial determinants and requires adenine and inositol for growth. The components of the heterokaryon are: